

Screening of Distilled Spirits for Thujone by Gas Chromatography-Mass Spectrometry

Scope:

Thujone is a monoterpene ketone consisting of two isomers, alpha and beta, that exist in varying ratios in different plants. Plants such as cedar leaf, sage, tansy, thyme, rosemary and wormwood (an ingredient found in certain flavored distilled spirits such as absinthe) are known to contain thujone. According to the FDA, alcoholic beverages must be thujone-free pursuant to 21 CFR 172.510. TTB has developed a gas chromatography/mass spectrometry (GCMS) screening method that may be used to quantitate total thujone (alpha + beta) in distilled spirits. The method has not been validated for wines, malt beverages, or nonbeverage products. Since menthol is added as an internal standard, all test samples should be verified for the absence of menthol.

The following is a synopsis of TTB's thujone screening method. The full screening method will be posted on TTB's website in December 2007.

Method:

Stock Solutions and Calibration Standards

500 ppm (-)- α -Thujone ($\geq 96\%$ pure) and 1000 ppm Menthol ($\geq 99\%$ pure) stock solutions are prepared in ethanol. Five calibration standards are prepared in 40% ethanol to contain: 1 ppm α -Thujone and 10 ppm Menthol; 2 ppm α -Thujone, 10 ppm Menthol; 5 ppm α -Thujone, 10 ppm Menthol; 20 ppm α -Thujone, 10 ppm Menthol; 50 ppm α -Thujone, 10 ppm Menthol.

Sample Preparation and GCMS analysis

5 mL of each calibration standard and 5 mL of testing sample(s) are transferred to separate glass test tubes. The testing samples are spiked additionally with menthol to 10 ppm. Upon the addition of 5 mL of saturated sodium chloride solution and 5 mL of methylene chloride, the solutions are extracted and the organic layer (bottom) is removed and placed into a GC vial for analysis.

N.B. If the alcohol content of the testing sample exceeds 45% ABV, the testing sample is diluted with DI water to 40% ABV and 5 mL of the resulting solution is extracted as described.

GCMS parameters: Scan acquisition mode; splitless injection; constant flow; DB-Wax or a similar 30 m capillary column.

Calculations and Reporting Data

Using the alpha-Thujone/Menthol ratio calibration curve, determine the total amount of thujone in the sample as the sum of alpha- and beta-thujone in ppm. The beta-thujone value is based on the alpha-thujone calibration curve. If the testing sample was diluted prior to extraction, the dilution must be factored. The GCMS screening method has a limit of quantitation of approximately 1 ppm.